

excess assay reagent and/or to wash reagents from the assay domain. Using this arrangement, a single device may be used to dispense assay reagents onto an assay domain (e.g., so as to cause localized immobilization of the assay reagent on the assay domain) and to wash excess assay reagent from the assay domain, these operations occurring without contamination of adjacent surfaces with the assay reagent. Optionally, an array of these devices is used to pattern and wash an array of assay domains.

[0176] The invention relates in part to assay cartridges. An assay cartridge of the invention incorporates one or more fluidic components such as compartments, wells, chambers, fluidic conduits, fluid ports/vents, valves, and the like and/or one or more detection components such as electrodes, electrode contacts, sensors (e.g., electrochemical sensors, fluid sensors, mass sensors, optical sensors, capacitive sensors, impedance sensors, optical waveguides, etc.), detection windows (e.g., windows configured to allow optical measurements on samples in the cartridge such as measurements of absorbance, light scattering, light refraction, light reflection, fluorescence, phosphorescence, chemiluminescence, electrochemiluminescence, etc), and the like. A cartridge may also comprise reagents for carrying out an assay such as binding reagents, detectable labels, sample processing reagents, wash solutions, buffers, etc. The reagents may be present in liquid form, solid form and/or immobilized on the surface of solid phase supports present in the cartridge. Certain preferred embodiments of the invention, comprise detection chambers having the electrode arrays and/or binding domains as described above (e.g., the electrode arrays described in **FIGS. 1-4**).

[0177] The fluidic components are preferably designed and incorporated into the cartridge body to form the fluidic network using certain predefined design guidelines. The design guidelines for each component can be dependent upon one or more factors such as, e.g., cartridge body design (i.e., single-piece body, multiple piece body, modular body, single read chamber, multiple read chamber, and the like), manufacturing process (e.g., injection molding, blow molding, hot stamping, casting, machining, etc.), materials (e.g., acrylic, PVDF, PET, polystyrene, polypropylene and the like), assay requirements (e.g., binding assay, competitive binding assay, single step assay, two-step assay, etc.), functional requirements (e.g., sample size, assay reagent volumes, detection technology, time-to-result, incubation, heating, mixing/agitating), safety/handling requirements (e.g., self-containment, regulatory approval, ease of use, etc.), and/or the like.

[0178] The skilled practitioner will be able to readily select materials suitable for the fabrication of the cartridges of the invention. Suitable materials include glass, ceramics, metals and/or plastics such as acrylic polymers (such as Lucite), acetal resins (such as Delrin), polyvinylidene fluoride (PVDF), polyethylene terephthalate (PET), polytetrafluoroethylene (e.g., Teflon), polystyrene, polypropylene, ABS, PEEK and the like. Preferably, the materials are inert to any solutions/reagents that will contact them during use or storage of the cartridge. In certain preferred embodiments, at least some portion of the cartridge is fabricated from transparent and/or translucent materials such as glass or acrylic polymer to provide windows that allow optical interrogation of fluids or surfaces inside the cartridge, e.g., for analysis of compositions within detection chambers of the cartridge or

for monitoring and controlling the movement of liquids through the fluidic networks defined within the cartridge.

[0179] One preferred embodiment of the invention is a cartridge that includes one or more sample chambers, one or more detection chambers (preferably, detection chambers adapted for use in ECL measurements as described above) and one or more waste chambers. The chambers are connected in series by fluid conduits so that a sample introduced into a sample chamber can be delivered into one or more detection chambers for analysis and then passed into one or more waste chambers for disposal. Preferably, this cartridge also includes one or more reagent chambers for storing liquid reagents, the reagent chambers connected via conduits to the other components so as to allow the introduction of the liquid reagents into specified sample or detection chambers. The cartridge may also include vent ports in fluidic communication with the sample, detection and/or waste chambers (directly or through vent conduits) so as to allow the equilibration of fluid in the chambers with the atmosphere or to allow for the directed movement of fluid into or out of a specified chamber by the application of positive or negative pressure.

[0180] In an alternative embodiment, a sample chamber and a waste chamber are both arranged upstream from a detection chamber having first and second inlet/outlet conduits (preferably, a detection chamber having an elongated shape, the inlet/outlet conduits being arranged at or near the opposite ends of the elongated dimension). The cartridge is configured to allow the introduction of sample into the detection chamber via the first inlet/outlet conduit and then the reversal of flow to direct the sample fluid back out the first inlet/outlet conduit and to the waste chamber. Preferably, a reagent chamber is located downstream of the detection chamber and the cartridge is configured to allow introduction of the reagent to the detection chamber via the second inlet/outlet conduit (i.e., in "reverse flow" relative to the introduction of sample). This arrangement is particularly well suited to measurements that suffer from strong sample interference, the reverse flow being especially efficient at washing residual sample from the detection chamber. This embodiment is especially useful in ECL-based assays for markers (e.g., cell wall markers of gram positive bacteria) in samples containing a nitrous acid-containing extraction buffer (see, e.g., the extraction methods and reagents disclosed in U.S. Provisional Patent Application 60/436,591, filed Dec. 26, 2002, entitled Methods Compositions and Kits for Biomarker Extraction, hereby incorporated by reference). One preferred embodiment of the invention uses a cartridge configured with a reverse flow wash to conduct an ECL binding assay for a panel of upper respiratory pathogens including streptococcal species and optionally other pathogens such as influenza A and B and RSV (preferably by employing an array of antibodies against markers of the pathogens, the array preferably being formed on one or more electrodes, most preferably an electrode array as described above and in **FIGS. 1-4**).

[0181] The reverse flow wash significantly reduces the detrimental effects of nitrous acid on ECL measurements. In preferred embodiments, the washing efficiency is such that the fraction of sample (or reagent) left in a detection chamber after a wash is less than 1/1000; more preferably less than 1/10,000, even more preferably less than 1/100,000